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STUDIES ON THE MINIMAL THRESHOLD OF THE DENTAL SIGN OF CHRONIC ENDEMIC FLUOROSIS (MOTTLED ENAMEL)

By H. TRENDLEY DEAN, *Dental Surgeon*, and ELIAS ELVOVE, *Senior Chemist*, *United States Public Health Service*

Since the publication of the results of three independent studies in 1931 (*1*), (*2*), (*3*), associating the presence of fluoride in the drinking water with the endemic hypoplasia of the permanent teeth known as mottled enamel, the question of what constitutes a permissible amount of fluoride in a domestic water supply frequently arises. Attempts to prevent the further development of this disease by removing the toxic amounts of fluorides present in the water is naturally dependent upon reliable and definite information concerning this permissible maximum, or minimal threshold.

For public health purposes we have arbitrarily defined the minimal threshold of fluoride concentration in a domestic water supply as the highest concentration of fluoride incapable of producing a definite degree of mottled enamel in as much as 10 percent [1] of the group examined. This group should consist of at least 25 children 9 years of age or older, who, since birth, had continuously used the water under investigation for both drinking and cooking. In cases where the examination of the first 25 children discloses an incidence of less than 75 percent it has been found desirable, if not necessary, to increase the number in the group to 50 or more. In other words, when more than 25 percent are diagnosed as "normal" or "questionable", the increase in the number of the group examined is necessary to compensate for fluctuations in sampling and their possible effect on the computation of the community mottled-enamel index. The thorough surveys thus far made, though limited in number, have indicated that, when an adequate number of children are examined, there is orderly uniformity in the group response to the fluoride concentration, with regard both to the incidence and the percentage distribution of severity, particularly the latter.

An attempt to determine the minimal threshold by a correlation of clinical observations with chemical findings is obviously unwarranted until certain variables intimately associated with the problem

[1] A community is given a "negative" mottled-enamel index when "less than 10 percent of the children show 'very mild' or more severe types of mottled enamel." (Ref. 8.)

are removed. Chief of these variables are (*a*) discontinuities in time of exposure, and (*b*) changes in the fluoride content of water supplies.

In considering the time variable it must be borne in mind that the causative factor of mottled enamel is probably operative during the entire period of calcification of the permanent teeth. An observed effect, therefore, may be the result of a comparatively low fluoride concentration operating during the entire period, or a somewhat higher concentration for a shorter period. In other words, in endemic areas where the common water supply contains fluoride in concentrations only slightly above the minimal threshold, the time of risk of exposure would probably have to be continuous with the period of calcification. On the other hand, in areas where water of a high fluoride concentration is used, its use during a fractional portion of the period of calcification is sufficient to produce the signs of mottled enamel. A careful analysis of the Bauxite survey (*4*) illustrates this point. Hence, in order to eliminate the influence of this variable, clinical observations were made only on those children whose histories showed that the water in question was used continuously since birth for both drinking and cooking. Breaks in continuity not exceeding a total of 30 days in any one calendar year are excepted.

The other major variable relates to changes in the fluoride content of the water supply. Any attempt to correlate clinical observations with a single fluoride determination of a common water supply associated with chronic endemic dental fluorosis clearly introduces a possible error. While it is true that fluctuations in mineral content of a water are least when the water is obtained from deep wells, nevertheless there may be changes in the physical set-up or source of the municipal water supply during the child's lifetime which would cast doubt upon any correlations which might be made. Stating it differently, the amount of flouride in a water sample taken at the time of a clinical survey of a group, for instance, of 12-year-old children, would be of little value unless the water supply had undergone no changes during a period concomitant with the life of the group of children examined.

Furthermore, when dealing with surface waters or shallow wells the seasonal and annual rainfall and other meteorological conditions introduce factors which would make it impossible to draw reliable conclusions from a single fluoride determination of the water in question. In this report the mean annual fluoride content, later referred to, represents an arithmetical mean of 12 consecutive monthly samples.

Eleven cities were chosen for study. In 6 of these mottled enamel was known to be endemic (*5*); 5 others, with as nearly as possible comparable conditions, but reported as nonendemic, were selected for "controls." These cities contained populations sufficiently large to

permit the necessary sifting and elimination of those children whose histories failed to show an exclusive use of the municipal water from birth. Six of the cities have populations (census of 1930) in excess of 10,000, the largest being more than 50,000. From each of these cities 12 consecutive monthly water samples of the municipal water supply were collected and forwarded to the National Institute of Health for fluoride determinations.

A report of a survey of four of these cities, prefaced by a description of their municipal water supply follows. These four cities are Colorado Springs, Colo.; Monmouth and Galesburg, Ill.; and Pueblo, Colo.

DESCRIPTION OF THE MUNICIPAL WATER SUPPLIES [2]

Colorado Springs, Colo.—The municipal water supply of Colorado Springs is obtained from surface sources; namely, melted snow from the south, west, and east slopes of Pike's Peak, and the east and west slopes of Mount Baldy. The water is stored in a system of seven mountain reservoirs, located at altitudes ranging from 9,300 to 12,000 feet. From this chain of reservoirs the water is conveyed through a transmission system to the settlers at Manitou, thence by gravity to three distributing reservoirs known as the "High Line", "Mesa No. 1", and "Mesa No. 2." These distributing reservoirs are located on a mesa just west of the city, and from these reservoirs begin the city distribution system and service mains. Water impounded in both the High Line and the Mesa Reservoirs is obtained from a common source and represents the type [3] of water used by the inhabitants of the city for at least the past 20 years.

Monmouth, Ill.—The municipal water supply of Monmouth is obtained from two wells 2,445 feet in depth. The first well was completed early in 1925, the second in 1926. Both wells obtain their water from the "Potsdam" stratum of the Cambrian sandstone. During the first 6 or 8 months of 1925 some water from the old wells in the St. Peter sandstone was added to that obtained from the first well, temporarily constituting a mixed supply. The percentage of the municipal water obtained from the old wells is not known, as the pumping records were not available. The second Cambrian well was completed early in 1926, and since that date all municipal water has been obtained from these two wells.

Both wells have 90 feet of 20-inch copperoid casing, 400 feet of 18-inch cast-iron casing, and 1,200 feet of 12-inch cast-iron casing. Water from the St. Peter sandstone, which is found between 1,100 and 1,250 feet in this locality, is apparently cased off. No strainers which permit mixture of water from higher strata are present in the casing. There has been practically no change in the water level of either well since installation. Water from these two wells is more than ample for municipal needs; one well is pumped during the day and the other during the night.

Galesburg, Ill.—The public water supply of Galesburg is obtained from two 2,414-foot wells drilled to the Cambrian sandstone. The first well was drilled in

[2] The description and data concerning these municipal water supplies were furnished by Messrs. B. B. MacReynolds, George M. Crow, H. O. Chambers, and D. P. Porter, superintendents of the Water Departments of Colorado Springs, Monmouth, Galesburg, and Pueblo, respectively.

[3] The only exception to the foregoing is that in the extreme southwestern panhandle-like projection of the westward city limits, a few lateral distribution mains lead off from the Bear Creek pipe line before ti connects with the transmission lines between Manitou and the Mesa Reservoirs. A small fraction of the population is served in this manner; the source, however, of the Bear Creek water is not far (½ mile) from Lake Moraine, one of the main reservoirs in the city mountain system. Because of this possible variable, children residing in this section of the city were not included in the group upon which the mottled enamel index was based.

1919, installed in 1920, and has been in continuous use since. In 1928 a second well of the same depth was completed. The casing record of both wells indicates that water from the St. Peter sandstone is completely cased off. Since 1928 these two wells have furnished practically all (more than 98 percent) of the water used by the population.

Between 1924 and 1928 the common water supply consisted of approximately 60 percent from the first "Potsdam" (2,414-foot) well and 40 percent from wells in the St. Peter sandstone. The latter wells (Central Fire Station and Brooks Street Station) were drilled in 1917 and 1918, and are 1,252 and 1,245 feet deep, respectively. Both of these latter wells are cased to the St. Peter sandstone so as to exclude water from higher levels. Water history prior to 1924 is omitted, as the time would be prior to the year of birth of the group of children examined and, therefore, not relevant.

Pueblo, Colo.—The municipal water supply of Pueblo is obtained from surface sources, namely, the Arkansas River. The city of Pueblo has two water systems, all of the city north of the Arkansas River being supplied by what is known as the "Pueblo Water Works, District No. 1", or "North Pueblo water supply", while that half of the city located south of the Arkansas River obtains its water from another system known as the "Pueblo Water Works, District No. 2", or "South Pueblo water supply." Both systems, however, obtain water from the Arkansas River.

The monthly water samples collected by the Pueblo City Health Department were all of the North Pueblo supply. Consequently the clinical examinations were limited to the children residing in that half of the city. A description of the North Pueblo supply follows:

Water is taken from the Arkansas River about 3 miles west of the city and diverted into reservoirs. In 1925 there were 6 reservoirs, and in 1928, 1 more was added. Treatment consists of preliminary sedimentation, coagulation with aluminum sulphate, followed by sedimentation and disinfection with ammonia-chlorine. Reservoirs nos. 1, 2, 3, and 4 are used for preliminary sedimentation, and reservoirs nos. 5, 6, and 7 for sedimentation after coagulation. The treatment does not include filtration. Prior to 1928 iron sulphate and lime were used as a coagulant, and prior to November 1931 chlorine was used without ammonia.

The North Pueblo water department has been obtaining water from this same source for the last 46 years. It might also be noted that between November 1933 and October 1934, the period during which samples were taken, there was an exceptionally low flow of the Arkansas River.

METHOD OF CLINICAL SURVEY [4]

In order to study clinical conditions in a group whose water history showed exclusive use of the water supply upon which the chemical determinations were made, examinations were limited to the 9-year-age group present in the school on the date of the examination. The selection of the 12-year group would have been more desirable, as practically all permanent teeth excepting the third molars are present in the mouth at that age. However, the present Monmouth water supply had been in use only 9 years at the time of the examination, and it was deemed advisable to limit the examination to the age group that had used the present supply exclusively since birth rather than

[4] The field surveys at Monmouth and Galesburg, Ill., were made during October 1934; those at Colorado Springs and Pueblo, Colo., during April 1935.

introduce an additional variable by selecting a higher age group. The same age group was then selected at Colorado Springs and Pueblo in order that the resultant data might be comparable. Brief reference will be made later to a variable present in the Galesburg water supply between 1924 and 1928.

The examinations were conducted in the following manner: First, all children who stated that they had not lived in the city continuously (30 days in a calendar year excepted) or had not used the municipal water supply exclusively were eliminated. The remainder were then called into the office of the school principal, one or two at a time, and further questioned as to their residence and water history. This cross-questioning often revealed breaks in continuous residence such as living a year or more in some nearby town or vacation trips in excess of 30 days. In addition there were a few instances of children who had lived in the community continuously but who had obtained their domestic water from other than the public supply. Children coming under these classifications were eliminated from further study.

If the questioning of the child elicited a history of continuous residence in the city and an exclusive use of the municipal water, the examination continued. The findings were recorded on a special card designed for mottled enamel surveys (fig. 1). The examinations were made in a good light with the child seated facing a window. Mouth mirrors, free of blemishes, and new explorers [5] were utilized in making the examination. In addition to mottled enamel, other defects of the enamel such as caries, present or past (fillings), pits and fissures, hypoplasias, etc., were recorded.

Following the examination, the home of each child was visited, and the information recorded in fig. 1 under "III. Water History" was carefully rechecked by an interview with the child's parent. This recheck revealed additional inaccuracies in residence or water supplies which the child either did not know or had forgotten. Under the conditions of this survey it was possible to find only approximately 20 percent in this age group who could demonstrate a continuous residence and an exclusive use of the city water during their lifetime. The results obtained are summarized in tables 1, 2, and 3.

In evaluating the Galesburg study, the variable in the city water supply between 1924 and 1928 must be borne in mind. However, it should be noted parenthetically that of the 15 individuals showing mottled enamel among the 39 rechecked cases, 11 of them had no erupted bicuspids. In the remaining 4 with bicuspids present, mottled enamel was observed in the bicuspids of each individual. A resurvey of Galesburg should be made 4 or 5 years later in order to determine the actual mottled enamel index.

[5] Explorers used were S8W no. 5, or equal.

UNITED STATES PUBLIC HEALTH SERVICE
DENTAL HYGIENE INVESTIGATIONS
MOTTLED ENAMEL STUDY

NAME OR NUMBER OF SCHOOL _____ EXAMINER _____

CASE NO. _____

STATE _____ NAME _____
LAST FIRST

CITY _____ AGE _____ SEX _____ COLOR _____
YEARS MONTHS

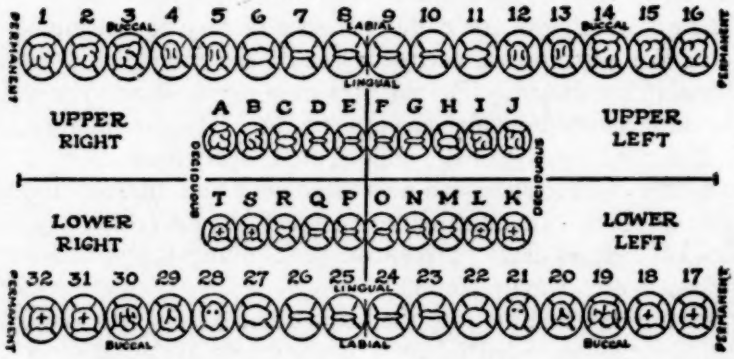
RURAL* _____ GRADE _____ DATE _____

* IF RURAL, NOTE DIRECTION AND MILES FROM NEAREST TOWN.

NOTE: I. AND II. MUST BE FILLED IN PRIOR TO III.

I. CLINICAL EXAMINATION. THE DIAGRAM MUST PRESENT DEFINITE INFORMATION CONCERNING EVERY TOOTH SHOWN IN IT. DRAW AN X THROUGH EACH MISSING TOOTH. DRAW A CIRCLE AROUND THE NUMBER OR LETTER OF EACH TOOTH THAT IS PRESENT AND NORMAL. OUTLINE AND FILL IN CAREFULLY ON TOOTH DESIGN THE AREA OF CARIES OR FILLING PRESENT. SYMBOLS TO BE PLACED UNDER APPROPRIATE TEETH:

UNERUPTED O; EXTRACTION INDICATED √; CROWN #; PONTIC £.
MOTTLED ENAMEL: WHITE OPAQUE =. BROWN STAIN +.



II. CLASSIFICATION OF MOTTLED ENAMEL DIAGNOSIS. FOR DESCRIPTION OF DEGREES OF SEVERITY, SEE J. A. D. A., AUGUST, 1934, OR P. H. R., MARCH 29, 1935. TO SUMMARIZE YOUR GENERAL IMPRESSION OF THE DEGREE OF SEVERITY, PLACE CHECK (√) IN ONE BOX:

NORMAL ☐ QUESTIONABLE ☐ VERY MILD ☐ MILD ☐
MODERATE ☐ MODERATELY SEVERE ☐ SEVERE ☐

FIGURE 1 (a).—Face of record form.

NOTE: I. AND II. MUST BE FILLED IN PRIOR TO III.

III. WATER HISTORY

RESIDENCE FROM BIRTH IN CHRONOLOGICAL ORDER *	DURATION (YRS.)	SOURCE OF DRINKING WATER **					
		MUNICIPAL	DEEP WELL	SHALLOW WELL	CISTERN	SPRING	OTHER
BIRTH PLACE							
2.							
3.							
4.							
5.							
6.							
7.							

WAS ABOVE HISTORY CONFIRMED BY INTERVIEW WITH CHILD'S PARENTS? YES ☐ NO ☐

NAMES OF BROTHERS OR SISTERS IN THE SAME SCHOOL: _____

REMARKS: _____

* IGNORE CHANGES IN A DURATION OF RESIDENCE LESS THAN THIRTY DAYS IN ONE CALENDAR YEAR.

** MUNICIPAL: DESCRIBE SEPARATELY AND IN DETAIL STATING TYPE AND HOW LONG PRESENT SUPPLY HAS BEEN USED. NOTE ALL CHANGES CONCOMITANT WITH LIFE OF AGE GROUP EXAMINED.
DEEP WELL: STATE DEPTH AND CASING; OBTAIN LOG IF AVAILABLE.
SHALLOW WELL: STATE WHETHER DUG OR DRIVEN AND APPROXIMATE DEPTH.
CISTERN: NOTE WHETHER CISTERN IS TIGHT OR LEAKY.
SPRING: STATE WHETHER HOT OR COLD AND TYPE OF GEOLOGICAL FORMATION THROUGH WHICH ISSUING, IF POSSIBLE.
OTHER: WRITE IN TYPE OF WATER SUPPLY, I.E., OPEN OR IRRIGATION DITCHES, CREEKS, ETC.

FIGURE 1 (b).—Back of record form.

TABLE 1.—*Summary of data with relation to continuity of residence and concomitant use of the municipal water*

	Colorado Springs	Monmouth	Galesburg	Pueblo (north of Arkansas River)
1. Number of grade public schools in city	10	4	9	8
2. Number of grade public schools in which examinations were held	5	4	4	4
3. Number of 9-year old pupils in attendance in (2) on date of examination	253	142	187	215
4. Number of pupils in (3) whose histories on questioning indicated constant residence and concomitant use of municipal water and who were examined	79	38	57	83
5. Percentage of age group present examined under (4)	31.2	26.7	30.5	[1] 38.6
6. Number of schedules eliminated by house-to-house recheck	25	9	18	34
7. Percentage of total present showing constant residence and water history	21.3	20.4	20.8	22.8

[1] This percentage is slightly higher than the others because of the dual water supply in Pueblo. Certain children were doubtful whether they had used the North Pueblo supply exclusively, but were nevertheless examined. When the house-to-house recheck revealed use of the South Pueblo water supply during some period of their life, these schedules were eliminated.

TABLE 2.—*Distribution according to severity* [1] *of those examined in (4), table 1*

City	Number of children	Normal	Questionable	Very mild	Mild	Moderate	Moderately severe	Severe	Incidence
		Number							
Colorado Springs	79	15	11	24	14	13	2	0	67.1
Monmouth	38	14	8	14	2	0	0	0	42.1
Galesburg	57	26	11	15	3	2	0	0	35.1
Pueblo	83	73	8	2	0	0	0	0	2.4
		Percent							
Colorado Springs	79	19.0	13.9	30.4	17.7	16.5	2.5	0	67.1
Monmouth	38	36.8	21.0	36.8	5.3	0	0	0	42.1
Galesburg	57	45.6	19.3	26.3	5.3	3.5	0	0	35.1
Pueblo	83	88.0	9.6	2.4	0	0	0	0	2.4

TABLE 3.—*Distribution according to severity* [1] *of those remaining after recheck (6), table 1*

City	Number of children	Normal	Questionable	Very mild	Mild	Moderate	Moderately severe	Severe	Incidence
		Number							
Colorado Springs	54	10	8	14	10	11	1	0	66.6
Monmouth	29	10	5	4	2	0	0	0	48.3
Galesburg	39	16	8	11	3	1	0	0	38.5
Pueblo	49	44	3	2	0	0	0	0	4.0
		Percent							
Colorado Springs	54	18.5	14.8	25.9	18.5	20.4	1.8	0	66.6
Monmouth	29	34.5	17.2	41.4	6.9	0	0	0	48.3
Galesburg	39	41.1	20.5	28.2	7.7	2.6	0	0	38.5
Pueblo	49	89.8	6.2	4.0	0	0	0	0	4.0

[1] For a detailed description of the various gradations of mottled enamel severity, see reference 8.

As has been noted previously, samples of the waters were obtained monthly. The fluoride content was estimated colorimetrically by means of the zirconium-alizarin reagent (*6*). The results obtained are given in table 4.

TABLE 4.—*Fluoride (F) content of monthly samples*

Month	Colorado Springs	Monmouth	Galesburg	Pueblo
	Parts per million			
1933				
November	2.6	1.6	1.8	0.6
December	2.9	1.6	1.8	.6
1934				
January	2.9	1.6	1.8	.6
February	3.0	1.7	1.9	.6
March	3.0	1.7	1.9	.6
April	3.0	1.7	1.9	.7
May	2.9	1.9	1.8	.6
June	2.3	1.7	1.9	.3
July	2.0	1.8	2.0	.5
August	1.8	1.8	1.9	.6
September	1.9	1.8	1.9	.6
October	2.0	1.6	1.8	.6
Mean annual fluoride content	2.5	1.7	1.86	0.57

While it may seem reasonable that the mottled enamel index will be found to depend entirely on the fluoride concentration of the drinking water, it is also possible that other constituents of the water may have some influence on the activity of the fluoride. For this reason we believe that a careful survey of a community for mottled enamel should, for the present at least, include also a chemical analysis of the drinking water for constituents other than fluoride.

Results of the chemical analyses of the waters [6] are given in table 5.

TABLE 5.—*Analyses of the waters used*

	Colorado Springs	Monmouth	Galesburg	Pueblo (north of Arkansas River)
	Parts per million			
Residue on evaporation	46.5	1,031.5	1,080.0	588.0
Loss on ignition	9.0	62.7	51.2	75.0
Fixed residue	37.5	968.8	1,028.8	513.0
Silica (SiO_2)	16.0	14.0	12.8	20.0
Iron (Fe)	.12	.18	.15	.14
Aluminum (Al)	.5	.3	.3	0
Calcium (Ca)	7.2	69.3	62.2	83.6
Magnesium (Mg)	.6	26.6	25.7	27.3
Sodium and potassium (calculated as Na)	5.4	249.9	288.8	54.9
Bicarbonate (HCO_3)	24.4	283.0	296.5	187.8
Sulfate (SO_4)	4.5	412.1	341.4	244.5
Nitrate (NO_3)	1.0	3.3	3.3	5.2
Chloride (Cl)	1.0	117.0	196.0	16.5
Fluoride (F)	2.0	1.8	1.9	.6
Phosphate (PO_4)	0	0	0	0
Boron (B)	0	.8	.9	0

[6] These samples of water from Monmouth and Galesburg, Ill., were received in September 1934, and those from Colorado Springs and Pueblo, Colo., were received in October 1934. Assistant Chemist C. G. Remsburg carried out the determinations other than fluoride and boron, using mostly the methods given in the Standard Methods of Water Analysis of the American Public Health Association. The boron determinations were made essentially by the method of Foote (*7*).

DISCUSSION

An analysis of the field data indicates the care that must be exercised in checking the continuity of residence and water history of the children under examination. As a result of checking the water histories as given by the child by a follow-up recheck with the parents, only about 20 percent of the children in the age group studied and present in the school on the day of the examination were found to have had an unbroken history of residence and constant use of the city water supply.

As a result of the house-to-house recheck, the mottled enamel incidence increased, however, only 6.2 percent in Monmouth, 3.4 percent in Galesburg, 1.6 percent in Pueblo, and decreased 0.5 percent in Colorado Springs. For the purpose of computing an approximate mottled enamel index of a community it would appear on the basis of these examinations that, while it is necessary to obtain accurate water histories by careful questioning, a house-to-house recheck may be omitted. The errors in the history as given by the child on individual questioning are apparently largely compensatory, not cumulative, and the effect on the figures representing the distribution of severity and incidence is comparatively small.

As was indicated in the introduction, the study undertaken included 6 cities where mottled enamel was known to be endemic, and 5 other cities with comparable conditions were included as "controls." The latter were considered as controls because, on the basis of available reports, they were apparently free of mottled enamel. Monmouth was one of the six cities which had been reported as having mottled enamel while Galesburg, because it was reported as apparently free of mottled enamel, was chosen as the "control" for Monmouth. But, as already indicated above, a careful survey shows that Galesburg does not differ materially from Monmouth, and both may be given the same mottled enamel index.

A cursory examination of the fluctuating fluoride content of the Colorado Springs municipal water indicates why correlation of clinical observations with a single fluoride determination should be made with extreme caution. In this instance, if the field study had been made in August the index based on the clinical observations would have been correlated with a fluoride content of 1.8 p. p. m. of fluoride, while if the same survey had been made in February, March, or April, the index would have been correlated with a fluoride content of 3.0 p. p. m.

In accordance with previously described means *(8)* of determining the mottled enamel index of a community, the application of this method to the percentage distribution of severity as listed in tables 2 and 3 indicates that the mottled enamel index of Colorado Springs, Monmouth, and Galesburg is "slight", and that of Pueblo is "negative."

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The authors are greatly indebted to Dr. O. R. Gillett, Dr. R. C. McMillan, Mr. F. M. Giddings, and Dr. W. E. Buck, city health officers of Colorado Springs, Monmouth, Galesburg, and Pueblo, respectively, for their assistance in collecting and forwarding the monthly water samples, and to Messrs. Hobart M. Corning, Roy Fetherston, O. O. Young, and J. A. Risley, superintendents of education of Colorado Springs, Monmouth, Galesburg, and Pueblo, respectively, for their cooperation and assistance in the field surveys.

SUMMARY

1. The "mottled enamel index" of Colorado Springs, Colo., is "slight." A milder type of mottled enamel is endemic in Monmouth and Galesburg, Ill.; the mottled enamel index of both communities is likewise "slight." In Pueblo, Colo., the index is "negative."

2. The mean annual fluoride (F) content, based on monthly examinations, of the municipal water of Colorado Springs, Colo., between November 1933 and October 1934 was close to 2.5 parts per million. The corresponding mean annual fluoride content of the municipal waters of Monmouth, Ill., Galesburg, Ill., and Pueblo, Colo., was close to 1.7, 1.8, and 0.6 parts per million, respectively.

3. In 4 groups of 9-year-old school children, numbering 142, 187, 215, and 253, respectively, in communities of a fairly stable population, only about 20 percent (minimum, 20.4; maximum, 22.8) were found to have a history of continuous residence and constant use of the municipal water supply from birth.

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A METHOD FOR THE STANDARDIZATION OF ANTIPNEUMOCOCCIC HORSE SERA AND CONCENTRATES [1]

By LLOYD D. FELTON, M. D., and HELENE J. STAHL, *Department of Preventive Medicine and Hygiene, Harvard Medical School*

The *in vitro* method for the standardization of antipneumococcic sera and concentrates here presented is an outgrowth of observations made during studies on serum antibodies. Preparations from immune serum, in which lessened precipitating activity with the soluble specific substance (SSS) of Heidelberger and Avery was noted, were frequently found to protect mice as well as did the original serum. This was found to be true of serum concentrates, in general, regardless of the method of concentration. In consequence, any method which relied upon the precipitating index as the criterion of serum antibody content gave disappointing results. However, it was noted that, although the precipitating titer of concentrated serum was relatively low, the function of the antibody to combine with SSS was not significantly diminished. This report describes the details of a method based on this combining power of antibody and SSS. A more complete discussion is given in another publication.

The test makes use of the principle of equivalent proportions in the combining of antibody with the specific polysaccharide of Heidelberger and Avery. The purity or degree of refinement of the SSS determines the actual number of antibody units which combines with a definite weight of the polysaccharide, but the combining ratio for any one preparation remains constant over a broad range of dilutions. Furthermore, within certain limits of concentration, the amount of antibody is in direct proportion to the amount of SSS used. This has been proved experimentally and may be shown graphically by plotting the equivalent units of antibody against the corresponding dilutions of SSS expressed in micrograms. Within the limits of polysaccharide concentrations from 10 to 50 micrograms per cubic centimeter, the resulting points fall in a straight line.

In equivalent mixtures of SSS and antibody, no excess of the polysaccharide is demonstrable by macroscopic methods. However, if there is too little antibody in the mixture to react with the total amount of SSS present, the unbound SSS may be precipitated by a second application of antibody. Therefore the "equivalent combining test" is carried out in the two steps which are briefly outlined as follows:

First step.—To a series of increasing serum dilutions is added a constant amount of SSS. The mixtures are thoroughly shaken and incubated for 2 hours at 37° C., after which they are stored over-

[1] This is one of a series of studies carried out in part under a grant from the Influenza Commission of the Metropolitan Life Insurance Co., and in part under a grant from the Pneumonia Fund of Harvard University.

night in the cold (4° C.) to insure a stable equilibrium. In the morning, the precipitate formed during this first step is separated from the supernatant fluid, which must be clear and free from particles for the second step.

Second step.—A definite amount of antibody ("indicating serum") is added to these supernatant fluids to indicate by the formation of a precipitate the mixtures in which unbound SSS is present. The first mixture in the series which contains no precipitate, or, in other words, no free SSS, is considered as the "equivalent combining mixture", and the percentage dilution of serum represented is called the "equivalent end point" of that serum.

A stoichiometric relationship exists between the unit value of a serum and its equivalent end point. However, the terms "equivalent end point" and "equivalent combining mixture", as employed in this test, do not represent absolute values, but rather values dependent upon the quality of the SSS preparation used and the sensitivity of the indicating serum. When conditions are uniform, and one polysaccharide preparation is used, the relative combining values of different sera may be translated into units of antibody per cubic centimeter by comparison with a similar value obtained with a standard serum of known unitage. The United States Government serum P 11 may be used as the standard serum, but owing to its low potency (300 type I units and 150 units of type II per cc) too much is required, considering its limited supply, for a satisfactory titration with a good preparation of the polysaccharide. A more economical and convenient standard is a serum containing at least 500 units of each type per cubic centimeter, which has been accurately standardized by the mouse-protection method with P 11 as a control.

The indicating serum is an important factor in the success of the test, for it determines the character of the precipitate by which the end point is chosen. The best for the purpose is a fresh specific immune horse serum of high precipitin titer used in an amount which gives a precipitate with a high dilution of SSS. This precipitate should be well formed, covering the bottom of the tube with a disk which can be seen by reflected light. While it is essential to add enough serum to indicate a small excess of free SSS, the addition of too much serum either altogether inhibits the formation of a precipitate (zone phenomenon), or so changes its character that it is translucent and sticky, and thus hard to read.

The optimum amount of an indicating serum is determined by experiment under the conditions of the test as follows: A uniform volume (0.2 cc) of decreasing dilutions of the serum (1:1, 1:2, 1:4, 1:8, or less, depending upon the serum) is added to 2-cc volumes of 1:1,000,000 SSS, which in reality is the smallest amount of free SSS ordinarily encountered in test mixtures. The mixtures are incubated for 2 hours at 37° C. and allowed to stand at room temperature for

2 hours. The precipitates are recorded, and the smallest amount of serum required to produce a satisfactory disk is chosen for use in equivalent combining tests.

The specific soluble substance of Heidelberger and Avery which is used in the test should be comparatively pure, showing approximately the following results of analysis: Precipitin titer of type I, 1:3,000,000, of type II, 1:5,000,000; nitrogen in type I, 4 percent, in type II, 0.3 to 1 percent; hydrolyzable sugars in type I, 26 to 30 percent, in type II, 40 to 50 percent. The dried material should dissolve completely at pH 7 to give a water-clear or slightly opalescent solution when made up 1:500 in physiological saline.

In addition to the requisites of the standard serum, the indicating serum and the SSS as stated above, there are certain considerations to be observed in the test procedure in order to minimize the experimental error.

The serum increment.—A uniform variation in the test dilutions of serum is desirable for accurate titration. Because of the difference in antibody content of individual sera and concentrates, a uniform dilution increment must be discussed in terms of units rather than volume. In general, reliable results are obtained when the increment is not less than 1 unit nor greater than 5 units. Actually, when the serum contains 300 units per cc, a variation of 5 percent by volume in a series of dilutions is equivalent to 15 units; but a 1 percent volume variation is equal to a 3-unit increment. Needless to say, a closer titration is obtained under the latter condition.

The range of serum dilutions.—In a series of dilutions ranging from 1 to 50 percent with a uniform increment of 1 percent, it is obvious that a greater mathematical error is inherent in the dilute portion of the series. For practical purposes, in order to minimize this error, any test which gives an equivalent end point in concentrations less than 10 percent is disregarded. The test is then repeated, making an initial dilution of the serum so that the end point will occur in the zone of dilutions above 10 percent.

The SSS dilutions.—If a good preparation of SSS is used, a 1:50,000 to 1:100,000 (20 to 10 micrograms per cc) will give an equivalent end point between 10 and 20 percent with a standard serum containing 300 to 500 units per cc. Such a dilution of the polysaccharide is most satisfactory for routine standardization purposes.

TECHNIQUE

Exact quantitative technique should be employed in the preparation of the test dilutions of both the SSS and the antibody. The following details are given in order to clarify the procedure.

1. *Preparation of SSS dilutions.*—Make the stock solution in a volumetric flask, from an accurately weighed sample of the dried material, dissolving it first in a small volume of saline with the addi-

tion of enough normal NaOH to maintain a neutral reaction and facilitate solution. Then add saline to complete the volume for a 1:500 dilution. Using a volumetric flask and a quantitative delivery pipette, prepare from this the dilution required for the test in such a quantity that not less than 1 cc of the 1:500 stock solution need be measured.

2. *Preparation of serum dilutions.*—Measure the physiological saline for the serum dilutions with a burette when more than 2 cc is needed, and with a 2-cc accurately calibrated serological pipette graduated in tenths, when less than 2 cc is needed. In measuring the serum for the initial dilution, draw it up to the line in a quantitative pipette calibrated to contain exactly the amount required. With a piece of cotton or gauze, remove the excess serum clinging to the outside of the pipette, and rinse the inside five times in the dilution being made. Use a 2-cc serological pipette graduated in tenths in making subsequent dilutions from this initial one. Make at least 1-cc volumes preferably 2 cc or more, of each dilution.

3. *Mixing the two components (first step).*—After the serum dilutions have been properly prepared, use quantitative pipettes to deliver 1-cc volumes into small agglutination tubes 4 inches by ⅝ inch, arranged in suitable racks. Then, with a quantitative delivery pipette, add 1 cc of the chosen dilution of SSS to each. Shake each tube to insure thorough mixing, incubate for 2 hours in a 37° C. water bath, in which the water extends half-way up to the level of the mixtures in the tubes. Store in the cold (4° C.) over night.

4. *Adding the indicating serum (second step).*—The next morning, without stirring up the precipitates, separate them from the supernatant fluids, by centrifugalization, preferably in the cold or at room temperature for 3 or 4 minutes. Drain the supernatant fluids into clean, clear agglutination tubes and to them add the indicating serum in the amount found to be optimal (see under indicating serum). Shake each tube thoroughly. Incubate at 37° C. for 2 hours. Do not disturb the precipitates when handling the racks after incubation, but allow them to settle for 2 hours at room temperature before recording the end point.

5. *Reading the end point.*—For consistent and reliable results, consider the end point as that dilution next in series to the one containing a well-formed disk precipitate. With some sera and some SSS preparations, the end point is extremely definite, i. e., of 2 adjacent tubes in a series, one contains a disk precipitate, the other contains not even a trace of flocculation. On the other hand, there are cases when the mixtures next in series to the last disk precipitate are not clear, but contain a fine sediment which becomes a swirl when the tube is shaken. This irregularity may be attributable to a multiplicity of antigens and (or) antibodies. Consequently, disregard these fine, dwindling

precipitates and record the tube following the last disk precipitate in the series as the equivalent end point.

6. *Calculating the unit value.*—The greater the unit value of any antibody preparation, the smaller is the volume required to combine equivalently with a given dilution of SSS. Hence an inverse proportion expresses the relationship between the antibody content of an immune serum and the percentage dilution that represents its equivalent end point. The unit value of unknown serum is found from the proportion $A:A'=B':B$, or $A=\frac{A'B'}{B}$, in which A equals the unit value of the unknown serum, A' the unit value of the standard serum, B the equivalent end point of the unknown, and B' the equivalent end point of the standard serum.

USUAL STANDARDIZATION PROCEDURE

The first step in standardizing an unknown serum is to find out its approximate value, whether it is of high or low potency. For this purpose either the precipitin or agglutinin titer affords a basis of estimation. A more useful measure, perhaps, is a combining test in which the serum dilutions are widely spaced in order to cover a broad range of concentrations. In this way, a rough estimation of unit value is made, from which a confirmatory test, using a smaller dilution variation, hence a smaller unit increment, is planned.

In the first column of the following table, possible end points of the preliminary test are given. In the next three columns the initial dilution, the dilution range, and the increment for the confirmatory test are shown for each case. It will be noted that the initial dilution is designed to insure a dilution range of from 20 to 10 percent whenever possible, and at the same time, a small unit value for the 1 percent increment.

Reference table

If the endpoint of the preliminary test is (percent)—	Then for the confirmatory test make—		
	(1) an initial serum dilution	(2) from which prepare a dilution series from—	(3) with an increment of—
		Percent	*Percent*
40	None	50 to 30	2
35	do	44 to 24	2
30	do	35 to 25	1
25	do	30 to 20	1
20	do	25 to 15	1
15	do	20 to 10	1
10	1:2	25 to 15	1
5	1:3	20 to 10	1
Less than 5	1:5 or 1:10	20 to 10	1

The figures in this table are valid when 10 to 20 percent end point represents a standard serum of 600 to 800 units per cc.

The following example is illustrative of the complete standardization procedure, performed in accordance with the principles outlined in this report.

Preliminary test.—Using the described quantitative technique, dilutions of an unknown bivalent antipneumococcic horse serum X 60 were prepared with a 5 percent increment over a range from 40 to 5 percent by volume. A 50 percent dilution was made by mixing equal volumes of serum and saline. The test dilutions were prepared in the following manner:

40 percent dilution=4.0 cc of 50 percent dilution+1.0 cc saline.
35 percent dilution=3.5 cc of 50 percent dilution+1.5 cc saline.
30 percent dilution=3.0 cc of 50 percent dilution+2.0 cc saline.
25 percent dilution=2.5 cc of 50 percent dilution+2.5 cc saline.
20 percent dilution=2.0 cc of 50 percent dilution+3.0 cc saline.
15 percent dilution=1.5 cc of 50 percent dilution+3.5 cc saline.
10 percent dilution=1.0 cc of 50 percent dilution+4.0 cc saline.
5 percent dilution=0.5 cc of 50 percent dilution+4.5 cc saline.

One cc of a 1:100,000 dilution of SSS was added to 1 cc of serum dilution and the test was completed in the prescribed manner. The protocol of this preliminary test is given below:

SSS 1:100,000	Percent dilutions of unknown serum					
	30	25	20	15	10	5
Type I	−	−	−•	+•	+	+
Type II	−	−	−	−	+	+

+ indicates disk precipitate.
• indicates preliminary end point.

Confirmatory test.—For this test the range and increment of the dilution series for serum X 60 were found in the reference table opposite the values for the preliminary end points. These were 20 and 15 percent for type I and type II antibody, respectively. Consequently, a dilution series which included concentrations from 25 to 10 percent of the serum by volume was prepared, with a 1 percent volume increment. From a 30 percent dilution (3 cc of serum+7 cc of saline), the dilutions from 25 to 20 percent were made as follows:

25 percent dilution=2.5 cc of 30 percent dilution+0.5 cc saline.
24 percent dilution=2.4 cc of 30 percent dilution+0.6 cc saline.
23 percent dilution=2.3 cc of 30 percent dilution+0.7 cc saline.
22 percent dilution=2.2 cc of 30 percent dilution+0.8 cc saline.
21 percent dilution=2.1 cc of 30 percent dilution+0.9 cc saline.

Likewise, from a 20 percent dilution (2 cc serum+8 cc saline) the series from 19 to 10 percent was made:

19 percent dilution=1.9 cc of 20 percent dilution+0.1 cc saline.
18 percent dilution=1.8 cc of 20 percent dilution+0.2 cc saline.
17 percent dilution=1.7 cc of 20 percent dilution+0.3 cc saline.
16 percent dilution=1.6 cc of 20 percent dilution+0.4 cc saline.
15 percent dilution=1.5 cc of 20 percent dilution+0.5 cc saline.
14 percent dilution=1.4 cc of 20 percent dilution+0.6 cc saline.
13 percent dilution=1.3 cc of 20 percent dilution+0.7 cc saline.
12 percent dilution=1.2 cc of 20 percent dilution+0.8 cc saline.
11 percent dilution=1.1 cc of 20 percent dilution+0.9 cc saline.
10 percent dilution=1.0 cc of 20 percent dilution+1.0 cc saline.

At the same time, the standard serum containing 500 units of each type per cubic centimeter was diluted in a similar way over a range from 20 to 10 percent of its concentration. The results of the completed test are recorded in the following table:

Serum	Type of SSS	Percent dilutions of serum										
		20	19	18	17	16	15	14	13	12	11	10
Standard	I	−	−	−	−	−*	+	+	+	+	+	+
X 60	I	−	−	−*	+	+	+	+	+	+	+	+
Standard	II	−	−	−	−	−	−	−*	+	+	+	+
X 60	II	−	−	−	−	−	−	−	−*	+	+	+

1:100,000 dilution of SSS used.
+indicates disk precipitate.
* indicates equivalent end point.

The experimental values for the equivalent end point of both the unknown and the standard serum, and the unit value of the latter, were substituted in the formula, $A=\frac{A'B'}{B}$. Thus for serum X60, the unit value is equivalent to: $A=\frac{500\times 16}{18}$, or 444 units of type I antibody, and $A=\frac{500\times 14}{13}$, or 538 units of type II antibody.

DISCUSSION

It is evident that the *in vitro* test just described applied to anti-pneumococcus horse sera and concentrates measures only the anti-SSS antibody which, of course, produces the usual immunological reactions. Antibodies to other antigens of the pneumococcus are known to be present in such sera, but so far they have not been shown to have any therapeutic value. The anti-C, or somatic antibody of Tillett, Goebel, and Avery (*1*), is present in what should be significant amounts. But it appears, especially when the study of Tillett and Francis (*2*) on pneumonia in man is taken into consideration, that this antibody has no curative effect. In other words, it is an antibody

developed in response to an antigen of the pneumococcus which bears no relationship to the pathogenesis of this organism to man.

On the other hand, according to the observations of Francis (*3*), Finland and Sutliff (*4*), Francis and Tillett (*5*), and Felton, Sutliff, and Steele (*6*), the proof that the SSS of Heidelberger and Avery is antigenic in that it stimulates antibody formation in man, and our unpublished results, as well, showing that the incidence of pneumonia is significantly lowered by its use as a prophylactic, emphasizes the importance of the anti-SSS antibody as a curative agent rather than other antibodies which might be present in antipneumococcus horse serum, but in relatively minute amounts. It is not to be inferred, however, that SSS is the essential or only antigen of the pneumococcus; for it is possible that other antigens may be found which, along with SSS, may stimulate antibodies capable of exerting greater curative action than anti-SSS antibody alone.

The importance of the anti-SSS antibody is further emphasized by the high degree of correlation between the results obtained by the mouse-protection test, using 40 mice to a dilution, and those of the combining equivalent test. The Pearson correlation coefficient of the results of this test compared with those of the mouse method in 40 sera and concentrates is, for type I, $r=0.90$, and for type II, $r=0.89$. This small correlation discrepancy may be due to experimental errors or may indicate the presence of an antibody other than the anti-SSS antibody which, for white mice, is curative. The facts of the case are not known. At any rate, with the antipneumococcus horse sera which are produced today, the curative value is for the most part measured by the anti-SSS antibody content.

In our opinion, when the material is to be used on human beings, no *in vitro* test can entirely supplant an *in vivo* test. For the last 3 years the combining equivalent procedure has been used to estimate the anti-SSS antibody content of sera and concentrates, and the results so obtained were checked by the usual mouse methods. This was done for two reasons: First, as an animal safety test, and, second, as a control on the *in vitro* method. Briefly, the mouse-protective titer was found to be equal to or higher than the equivalent combining titer in all tests run.

In consequence, the following procedure is being used and advocated for the standardization of antipneumococcus horse sera and concentrates for therapeutic purposes: First, the anti-SSS antibody content is estimated by the equivalent combining technique. This is then checked by the mouse method. For this purpose a single dilution each of the control and the unknown serum is tested, using only 10 mice for a dilution. The culture employed is of such virulence that 0.5 cc of a 1:200 dilution (or greater) of a 6- to 8-hour broth culture gives a satisfactory end point with the control serum. In case of the United

States Government serum P 11, the type I culture dose is used such that 3 to 7 mice survive when injected simultaneously with the culture and 0.5 cc of 1:300 dilution of this serum; the type II culture is used so that 3 to 7 mice survive with 0.5 cc of 1:150 serum dilution. Having established the satisfactory dose of culture for the end point of the control serum, the unknown can be checked. For example, if an unknown serum is estimated to contain 1,100 units per cc by the equivalent combining test, then 0.5 cc of a 1:1,100 dilution is injected in 10 mice along with the appropriate dose of culture. The control serum is injected as indicated above.

In the interpretation of the results of the mouse test after a 96-hour period there are three possibilities: First, if 3 to 7 mice survive in the case of the standard serum, and 7 to 3 in the unknown, the test is valid. Second, if all mice die with the control and 3 to 7 survive with the unknown, the test is also valid. Third, if less than three die with the control, regardless of what happens in case of the unknown serum, the check is not satisfactory. Accordingly, in the event of the first or second possibility, the unit value of the example given would be 1,100 units per cc. The results denoted in the third possibility indicate the need of a repetition of the test.

It should be understood that this method of checking with a small number of mice does not directly estimate mouse-protective units. It serves as an animal safety test and, in addition, proves that the anti-SSS antibody is active. In reality, the protective characteristic of antipneumococcus horse serum is a function of the anti-SSS antibody. Consequently, as long as this antibody is not altered, the combining equivalent test measures the protective units of both sera and concentrates. As a matter of fact, given a single sample of polysaccharide, the unit of antibody may be established as that amount which combines with a specified amount of SSS. For example, in one sample of type I SSS used in this study, 10 micrograms were equivalent to 54 protective units; and with type II, 10 micrograms were equivalent to 22 protective units. It would thus seem possible to estimate directly the protective unit value of an unknown serum by a standard SSS preparation rather than by a standard or control serum. The advantage of such a procedure is obvious, inasmuch as the polysaccharide is a more stable substance than the serum antibody.

REFERENCES

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A REPORT ON THE FUMIGATION OF A LOADED VESSEL

The following account of an interesting experience with the fumigation of a loaded vessel has been reported by Senior Surgeon C. L. Williams.

"The American S. S. *Delvalle* arrived in New Orleans on September 4 from Buenos Aires and other east-coast South American ports, loaded to the hatch coamings with birdseed, corn meal, corn, corned beef, tallow, and coffee, most of the cargo in packages, except the corn in bulk loaded in no. 5 hold. Inspection showed the presence of rats in no. 2 hold, the shelter deck over no. 3 hold, and in no. 5 hold, an estimate of 8 rats being made. In consequence, the ship was fumigated before discharge of cargo.

"At the time of fumigation there appeared to be no unusual features of construction or fumigation. On account of the full load, the gas in all holds was introduced at least in part through the ventilators, particular attention being paid to introducing proportionate dosages to the lower levels as well as to the 'tween decks. It was thought that in no. 3 hold, as well as in the others, all levels had been properly gassed. All holds were fumigated with liquid HCN, using the air-jet sprayer.[1]

"On the following day, while the ship was being unloaded, it became apparent to the inspector sent to determine the efficacy of the fumigation that there were still live rats in no. 3 'tween deck. He made a careful investigation and discovered that, on account of the peculiar construction of no. 3 hold, the 'tween deck was not reached by any ventilator; and since at the time of fumigation the shelter deck was full of cargo, completely covering up the 'tween-deck hatch, it was quite apparent that practically no gas had ever reached this location. The inspector promptly set traps in the 'tween deck, and next morning two more rats were recovered. No additional rats were trapped there later, nor were any rats trapped in any other part of the ship, although about 20 rat traps were set therein. Besides these 5 rats trapped, 8 rats were found dead—1 in the shelter deck of no. 3 hold, 1 in the forepeak, and 6 in no. 2 hold. The latter group consisted of an adult and 5 infant rats in a casing near the bottom of the ship.

"This instance is reported as an example both of effectiveness of loaded fumigation when carefully carried out and of the possibilities of missing certain compartments when the construction of the ship is not thoroughly understood or when the compartments are completely covered by cargo without access through ventilators."

[1] For a description of this sprayer see the Pub. Health Rep., vol. 46, no. 30, July 24, 1931, pp. 1755-1761.—Ed.

DEATHS DURING WEEK ENDED NOV. 16, 1935

[From the Weekly Health Index, issued by the Bureau of the Census, Department of Commerce]

	Week ended Nov. 16, 1935	Corresponding week, 1934
Data from 86 large cities of the United States:		
Total deaths	7,727	7,831
Deaths per 1,000 population, annual basis	10.8	10.9
Deaths under 1 year of age	495	571
Deaths under 1 year of age per 1,000 estimated live births	45	53
Deaths per 1,000 population, annual basis, first 46 weeks of year	11.3	11.3
Data from industrial insurance companies:		
Policies in force	67,721,419	67,041,531
Number of death claims	10,254	11,357
Death claims per 1,000 policies in force, annual rate	7.9	8.8
Death claims per 1,000 policies, first 46 weeks of year, annual rate	9.5	9.8

PREVALENCE OF DISEASE

No health department, State or local, can effectively prevent or control disease without knowledge of when, where, and under what conditions cases are occurring

UNITED STATES

CURRENT WEEKLY STATE REPORTS

These reports are preliminary, and the figures are subject to change when later returns are received by the State health officers

Reports for Weeks Ended Nov. 23, 1935, and Nov. 24, 1934

Cases of certain communicable diseases reported by telgraph by State health officers for weeks ended Nov. 23, 1935, and Nov. 24, 1934

Division and State	Diphtheria		Influenza		Measles		Meningococcus meningitis	
	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934
New England States:								
Maine	----	1	----	----	121	13	0	0
New Hampshire	1	----	----	----	----	5	0	0
Vermont	----	4	----	----	41	2	0	0
Massachusetts	7	17	----	----	62	83	2	0
Rhode Island	----	2	----	----	32	----	0	0
Connecticut	1	1	2	1	55	222	1	0
Middle Atlantic States:								
New York	32	45	[1] 4	[1] 41	481	1,078	8	5
New Jersey	25	32	8	26	23	49	0	1
Pennsylvania	60	56	----	----	133	632	8	2
East North Central States:								
Ohio	90	97	11	6	115	101	1	1
Indiana	89	53	15	43	7	149	0	1
Illinois	87	76	14	16	22	333	4	5
Michigan	23	12	2	3	37	47	6	1
Wisconsin	5	9	40	3	76	212	0	0
West North Central States:								
Minnesota	7	4	1	1	41	220	1	0
Iowa	28	4	----	----	7	277	1	0
Missouri	68	60	86	41	17	71	3	4
North Dakota	2	10	13	----	8	29	0	0
South Dakota	2	4	1	----	9	24	0	0
Nebraska	7	14	----	----	3	7	2	0
Kansas	16	15	12	----	11	83	0	1
South Atlantic States:								
Delaware	2	----	1	1	64	----	0	0
Maryland [2]	16	14	9	8	6	35	3	0
District of Columbia	23	14	2	----	2	1	2	0
Virginia [2]	68	104	----	----	23	140	6	2
West Virginia	48	60	25	33	18	161	0	3
North Carolina	78	74	10	10	37	107	0	0
South Carolina	8	10	163	267	5	3	0	0
Georgia [4]	27	52	7	----	2	----	1	0
Florida	12	25	2	----	1	10	0	0
East South Central States:								
Kentucky	61	79	17	32	5	332	2	0
Tennessee	60	62	38	68	7	22	3	0
Alabama [4]	37	37	15	51	12	57	1	1
Mississippi [2]	23	26	----	----	----	----	0	1

See footnotes at end of table.

Cases of certain communicable diseases reported by telegraph by State health officers for weeks ended Nov. 23, 1935, and Nov. 24, 1934—Continued

Division and State	Diphtheria		Influenza		Measles		Meningococcus meningitis	
	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934
West South Central States:								
Arkansas	28	16	44	19			0	0
Louisiana	24	30	4	5	8	8	4	0
Oklahoma [3]	23	16	51	27	2	6	5	0
Texas [4]	153	93	147	81	3	6	0	0
Mountain States:								
Montana	4	11	11	3	19	53	0	0
Idaho		1	2	3	14	11	0	0
Wyoming		3			3	4	0	0
Colorado	10	9			5	150	1	0
New Mexico	5	5	4	4		45	3	0
Arizona	5	5	36	15	1	2	1	0
Utah [2]					1	12	0	0
Pacific States:								
Washington		12			87	31	1	0
Oregon	2		23	42	264	15	0	2
California	62	42	44	32	204	148	4	0
Total	1,329	1,316	864	882	2,094	4,996	74	30
First 47 weeks of year	33,031	35,331	111,757	57,392	709,423	692,526	5,075	2,059

Division and State	Poliomyelitis		Scarlet fever		Smallpox		Typhoid fever	
	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934
New England States:								
Maine	0	0	24	26	0	0	2	5
New Hampshire	0	0	6	6	0	0	1	0
Vermont	0	0	5	14	0	0	1	1
Massachusetts	10	0	192	182	0	0	1	1
Rhode Island	2	0	9	17	0	0	0	0
Connecticut	6	0	54	45	0	0	1	2
Middle Atlantic States:								
New York	28	1	395	390	0	0	13	16
New Jersey	4	0	79	131	0	0	11	9
Pennsylvania	13	0	397	408	0	0	15	14
East North Central States:								
Ohio	1	0	252	447	0	1	4	5
Indiana	0	1	189	179	2	1	3	1
Illinois	7	3	485	590	2	1	12	24
Michigan	8	4	222	286	0	0	4	9
Wisconsin	2	0	352	433	0	15	3	1
West North Central States:								4
Minnesota	4	2	257	91	0	8	3	0
Iowa	2	1	116	63	2	0	3	5
Missouri	1	0	150	112	2	2	3	20
North Dakota	0	0	31	43	6	1	0	3
South Dakota	0	0	73	21	8	23	1	0
Nebraska	0	0	85	34	48	0	0	0
Kansas	0	1	127	53	11	1	6	4
South Atlantic States:								
Delaware	0	0	10	11	0	0	0	3
Maryland [2]	5	2	71	115	0	0	15	0
District of Columbia	1	0	2	24	0	0	0	0
Virginia [3]	1	1	51	162	0	0	13	14
West Virginia	0	2	119	177	2	1	5	11
North Carolina	7	0	76	123	0	0	4	2
South Carolina	1	0	2	6	0	0	2	3
Georgia [4]	0	0	23	23	0	0	2	2
Florida	0	0	7	8	0	0	2	3
East South Central States:								
Kentucky	4	1	75	84	0	0	7	24
Tennessee	6	1	74	103	5	0	4	15
Alabama [4]	0	2	12	43	0	1	2	13
Mississippi [3]	0	0	28	29	1	0	8	4

See footnotes at end of table.

Cases of certain communicable diseases reported by telegraph by State health officers for weeks ended Nov. 23, 1935, and Nov. 24, 1934—Continued

Division and State	Poliomyelitis		Scarlet fever		Smallpox		Typhoid fever	
	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934	Week ended Nov. 23, 1935	Week ended Nov. 24, 1934
West South Central States:								
Arkansas	0	0	13	13	2	0	2	18
Louisiana	0	1	15	21	0	1	9	12
Oklahoma [5]	1	0	20	22	0	0	11	33
Texas [4]	0	3	66	64	0	0	31	35
Mountain States:								
Montana	1	0	116	18	40	0	0	1
Idaho	0	0	34	13	0	0	2	1
Wyoming	0	0	76	12	0	3	0	0
Colorado	0	0	85	208	1	2	1	3
New Mexico	1	0	25	25	0	1	11	29
Arizona	1	1	32	36	0	0	1	9
Utah [3]	0	0	105	39	0	0	0	0
Pacific States:								
Washington	0	7	80	60	37	24	0	3
Oregon	6	2	64	59	0	0	4	1
California	14	23	245	198	7	2	7	11
Total	137	59	5,026	5,276	182	88	230	369
First 47 weeks of year	10,406	7,021	221,889	189,104	6,495	4,500	16,501	19,672

[1] New York City only.
[2] Week ended earlier than Saturday.
[3] Rocky Mountain spotted fever, week ended Nov. 23, 1935, Virginia, 1 case.
[4] Typhus fever, week ended Nov. 23, 1935, 18 cases, as follows: Georgia, 4; Alabama, 8; Texas, 6.
[5] Exclusive of Oklahoma City and Tulsa.

SUMMARY OF MONTHLY REPORTS FROM STATES

The following summary of cases reported monthly by States is published weekly and covers only those States from which reports are received during the current week.

State	Meningococcus meningitis	Diphtheria	Influenza	Malaria	Measles	Pellagra	Poliomyelitis	Scarlet fever	Smallpox	Typhoid fever
October 1935										
Alabama	2	177	76	858	16	28	3	60	0	24
Idaho			4		15		0	175	1	15
Illinois	19	294	61	34	61	1	58	1,692	7	92
Kansas	4	69	3	4	12		8	341	10	28
Louisiana	4	110	31	526	26	18	10	65	1	72
Maryland	19	58	21	18	34	1	15	299	0	76
Massachusetts	13	35		4	179		236	552	0	13
New York	32	113		4	729		285	1,218	0	63
Oklahoma [1]	8	71	102	272	6	11		68	2	53
Oregon		4	72	15	459		9	165	0	8
South Dakota	5	26	4	1	31		6	147	36	5
Texas	4	440	409	3,233	27	49	9	200	5	117
Virginia	9	280	255	124	29	6	19	251	0	57
West Virginia	4	240	55		24		1	523	1	48

[1] Exclusive of Oklahoma City and Tulsa.

Summary of monthly reports from States—Continued

October 1935

	Cases
Anthrax:	
Illinois	1
New York	1
Chicken pox:	
Alabama	50
Idaho	31
Illinois	773
Kansas	235
Louisiana	2
Maryland	115
Massachusetts	351
New York	976
Oklahoma [1]	11
Oregon	56
South Dakota	65
Texas	55
Virginia	100
West Virginia	90
Dengue:	
Alabama	1
Texas	11
Diarrhea:	
Maryland	39
Dysentery:	
Illinois (amoebic)	13
Illinois (bacillary)	28
Kansas (amoebic)	1
Kansas (bacillary)	5
Louisiana (amoebic)	7
Louisiana (bacillary)	3
Maryland	34
Massachusetts (amoebic)	1
Massachusetts (bacillary)	2
New York (amoebic)	3
New York (bacillary)	150
Oklahoma [1]	11
Oregon (amoebic)	1
Texas (amoebic)	1
Texas (bacillary)	31
Virginia (diarrhea included)	127
West Virginia	7
Epidemic encephalitis:	
Alabama	1
Illinois	12
Kansas	1
Louisiana	1
Maryland	1
Massachusetts	13
New York	12
Texas	1
Virginia	1
West Virginia	1
German measles:	
Alabama	1
Illinois	31
Kansas	13
Maryland	25
Massachusetts	39
New York	123
Hookworm disease:	
Illinois	1
Louisiana	13
Oklahoma [1]	3

October 1935—Con.

	Cases
Impetigo contagiosa:	
Illinois	27
Kansas	4
Maryland	60
Oklahoma [1]	10
Oregon	128
Lead poisoning:	
Illinois	3
Massachusetts	1
Leprosy:	
Louisiana	1
Milk sickness:	
Illinois	1
Mumps:	
Alabama	12
Idaho	19
Illinois	296
Kansas	116
Louisiana	1
Maryland	54
Massachusetts	333
Oklahoma [1]	14
Oregon	65
South Dakota	64
Texas	205
Virginia	54
West Virginia	5
Ophthalmia neonatorum:	
Illinois	4
New York	5
Oregon	1
South Dakota	1
Virginia	2
Paratyphoid fever:	
Illinois	2
Maryland	1
New York	6
Oregon	1
Texas	1
Virginia	1
Puerperal septicemia:	
Illinois	2
Rabies in animals:	
Alabama	75
Illinois	15
Kansas	9
Louisiana	24
Maryland	2
Massachusetts	9
New York [2]	3
Oregon	6
Texas	26
Rabies in man:	
Alabama	1
West Virginia	1
Scabies:	
Idaho	1
Maryland	2
Oregon	47
Septic sore throat:	
Illinois	6
Kansas	7
Louisiana	1
Maryland	14
Massachusetts	8

October 1935—Con.

	Cases
Septic sore throat—Contd.	
New York	46
Oklahoma [1]	25
Oregon	11
Virginia	8
Tetanus:	
Alabama	7
Illinois	5
Louisiana	6
Maryland	2
Massachusetts	2
New York	4
Virginia	2
Trachoma:	
Illinois	53
Massachusetts	5
Oklahoma [1]	18
South Dakota	2
Trichinosis:	
Illinois	5
Massachusetts	3
New York	6
Tularaemia:	
Illinois	1
Kansas	1
Louisiana	2
Virginia	6
Typhus fever:	
Alabama	25
Louisiana	1
New York	6
Texas	28
Undulant fever:	
Alabama	6
Illinois	11
Kansas	11
Louisiana	4
Maryland	5
Massachusetts	8
New York	15
Oklahoma [1]	1
South Dakota	1
Virginia	2
Vincent's infection:	
Illinois	19
Kansas	2
Maryland	17
New York [2]	79
Oklahoma [1]	5
Oregon	20
Whooping cough:	
Alabama	41
Idaho	5
Illinois	656
Kansas	96
Louisiana	54
Maryland	88
Massachusetts	286
New York	1,217
Oklahoma [1]	24
Oregon	6
South Dakota	24
Texas	80
Virginia	25
West Virginia	33

[1] Exclusive of Oklahoma City and Tulsa.

[2] Exclusive of New York City.

WEEKLY REPORTS FROM CITIES

City reports for week ended Nov. 16, 1935

This table summarizes the reports received weekly from a selected list of 140 cities for the purpose of showing a cross section of the current urban incidence of the communicable diseases listed in the table. Weekly reports are received from about 700 cities, from which the data are tabulated and filed for reference.

State and city	Diphtheria cases	Influenza		Measles cases	Pneumonia deaths	Scarlet fever cases	Smallpox cases	Tuberculosis deaths	Typhoid fever cases	Whooping cough cases	Deaths, all causes
		Cases	Deaths								
Maine:											
Portland	0	----	0	0	1	0	0	0	0	14	24
New Hampshire:											
Concord	0	----	0	0	0	2	0	0	0	0	12
Manchester	0	----	0	0	2	1	0	1	0	0	18
Nashua	0	----	----	0	----	0	0	----	0	0	----
Vermont:											
Barre	1	----	0	0	0	0	0	0	0	0	3
Burlington	0	----	0	0	0	0	0	0	4	0	3
Massachusetts:											
Boston	2	----	1	14	18	48	0	7	0	4	195
Fall River	1	----	0	0	0	1	0	1	0	0	16
Springfield	0	----	0	0	1	3	0	1	0	7	47
Worcester	0	----	0	1	3	21	0	2	0	4	51
Rhode Island:											
Pawtucket	0	----	0	0	0	0	0	0	0	0	12
Providence	1	----	0	0	3	5	0	0	0	3	60
Connecticut:											
Bridgeport	0	----	0	0	0	2	0	0	0	0	21
Hartford	0	----	0	1	1	6	0	1	0	8	----
New Haven	0	----	1	0	2	0	0	2	0	6	54
New York:											
Buffalo	1	----	0	12	15	39	0	5	0	10	125
New York	21	7	4	85	87	69	0	67	3	99	1,321
Rochester	0	----	0	2	2	3	0	2	0	4	52
Syracuse	0	----	0	0	1	3	0	0	0	18	43
New Jersey:											
Camden	1	1	0	1	3	2	0	0	0	0	19
Newark	0	2	0	1	2	18	0	10	0	40	80
Trenton	0	----	0	0	2	4	0	1	2	4	28
Pennsylvania:											
Philadelphia	10	1	1	27	18	88	0	22	2	68	425
Pittsburgh	4	7	3	4	18	67	0	3	0	20	165
Reading	0	----	0	3	0	3	0	0	0	0	23
Scranton	0	----	----	2	----	7	0	----	0	0	----
Ohio:											
Cincinnati	7	----	0	0	3	15	0	11	1	1	125
Cleveland	7	21	1	0	11	22	0	14	0	53	176
Columbus	4	1	1	0	2	11	0	4	0	1	67
Toledo	2	----	0	3	2	11	0	4	0	10	70
Indiana:											
Anderson	0	----	0	0	0	1	0	1	0	3	5
Fort Wayne	15	----	0	0	2	13	1	0	0	0	32
Indianapolis	10	----	1	2	11	15	0	2	0	12	107
Muncie	0	----	0	3	2	0	0	0	0	0	5
South Bend	6	----	0	1	3	1	0	0	0	1	16
Terre Haute	2	----	0	0	0	2	0	0	0	0	16
Illinois:											
Alton	4	----	0	0	0	7	0	0	0	0	10
Chicago	17	8	7	7	44	159	0	45	2	97	661
Elgin	1	----	0	0	1	4	0	0	0	0	13
Moline	0	----	0	0	1	1	0	0	0	0	7
Springfield	0	----	0	1	1	4	0	0	0	0	18
Michigan:											
Detroit	11	2	1	3	13	48	0	9	1	117	233
Flint	3	----	0	1	4	11	0	1	0	5	23
Grand Rapids	0	----	0	1	3	4	0	2	0	2	35
Wisconsin:											
Kenosha	0	----	0	1	0	8	0	0	0	4	4
Milwaukee	0	----	0	3	8	31	0	3	0	46	108
Racine	1	----	0	0	2	23	0	2	0	16	17
Superior	0	----	0	0	0	3	0	0	0	0	5
Minnesota:											
Duluth	0	----	0	2	1	1	0	0	0	9	15
Minneapolis	2	----	0	3	8	67	0	1	1	9	88
St. Paul	1	1	1	2	3	28	0	5	0	0	56

City reports for week ended Nov. 16, 1935—Continued

State and city	Diphtheria cases	Influenza		Measles cases	Pneumonia deaths	Scarlet fever cases	Smallpox cases	Tuberculosis deaths	Typhoid fever cases	Whooping cough cases	Deaths, all causes
		Cases	Deaths								
Iowa:											
Cedar Rapids	1		0	0	0	0	0	0	0	1	
Davenport	0			0		4	0		0	1	
Des Moines	1			1		5	0		0	0	37
Sioux City	3			1		8	0		0	0	
Waterloo	5			1		2	0		0	1	
Missouri:											
Kansas City	11		1	1	7	4	0	0	0	0	91
St. Joseph	7		0	1	3	2	0	1	0	0	45
St. Louis	10		1	0	5	26	0	6	1	5	155
North Dakota:											
Fargo	0		0	0	0	3	2	0	0	0	7
Grand Forks	0			0		0	0		0	0	
Minot	0		0	0	0	2	0	0	0	0	4
South Dakota:											
Aberdeen	0			0		0	0		0	0	
Nebraska:											
Omaha	10		1	1	3	56	0	1	0	0	46
Kansas:											
Lawrence	0		0	0	0	0	0	0	0	0	5
Topeka	0		0	0	5	10	0	0	0	3	13
Wichita	0		0	0	1	6	0	0	0	0	23
Delaware:											
Wilmington	0		0	0	3	0	0	1	0	0	22
Maryland:											
Baltimore	6	2	2	1	19	31	0	12	3	20	208
Cumberland	2		0	0	0	2	0	0	0	0	14
Frederick	0		0	0	0	1	0	0	0	0	1
District of Columbia:											
Washington	15	1	1	1	8	8	0	6	1	2	171
Virginia:											
Lynchburg	4		0	1	1	1	0	1	0	6	9
Norfolk	3		0	0	5	0	0	0	0	2	44
Richmond	3		0	0	5	5	0	2	1	0	51
Roanoke	0		0	1	0	5	0	1	1	0	17
West Virginia:											
Charleston	5		0	0	0	6	0	1	0	0	23
Huntington	2			0		4	0		0	0	
Wheeling	3		0	0	1	5	0	0	0	6	18
North Carolina:											
Gastonia	1		0	0	0	0	0	0	0	0	2
Raleigh											
Wilmington	0		0	0	0	0	0	0	0	0	10
Winston-Salem	1		0	2	2	5	0	0	0	0	15
South Carolina:											
Charleston	0	8	0	0	1	1	0	2	1	0	17
Columbia	0		0	0	0	0	0	0	0	0	4
Florence	0		0	0	1	0	0	0	0	0	9
Greenville	0		0	0	1	0	0	0	0	0	11
Georgia:											
Atlanta	8	7	0	0	5	14	0	5	0	1	75
Brunswick	0		0	0	0	1	0	0	0	3	2
Savannah	4	3	0	1	3	4	0	2	0	1	27
Florida:											
Miami	1	1	0	0	0	1	0	2	1	4	20
Tampa	6	1	0	0	1	2	0	1	0	0	25
Kentucky:											
Ashland	4			0		0	0		0	0	
Covington	3		0	0	1	2	0	0	0	0	14
Lexington	2		0	0	2	2	0	1	4	0	21
Louisville	9	4	1	2	4	10	0	0	2	1	59
Tennessee:											
Knoxville	6		1	0	1	2	0	0	1	0	21
Memphis	5		0	0	1	4	0	1	0	4	88
Nashville	6		0	0	2	4	0	2	0	0	45
Alabama:											
Birmingham	2	2	0	0	5	1	0	4	1	0	57
Mobile	5		0	0	2	1	0	0	0	0	14
Montgomery	1			1		2	0		0	0	
Arkansas:											
Fort Smith	1			0		0	0		0	0	
Little Rock	0		0	0	5	2	0	0	0	0	5
Louisiana:											
Lake Charles	0		0	0	0	1	0	0	0	0	6
New Orleans	13		0	0	11	2	0	9	0	10	141
Shreveport	2		0	0	5	2	0	7	1	0	43

City reports for week ended Nov. 16, 1935—Continued

State and city	Diphtheria cases	Influenza		Measles cases	Pneumonia deaths	Scarlet fever cases	Smallpox cases	Tuberculosis deaths	Typhoid fever cases	Whooping cough cases	Deaths, all causes
		Cases	Deaths								
Oklahoma:											
Oklahoma City	0	14	0	0	3	2	0	1	0	2	26
Tulsa	----	----	----	----	----	----	----	----	----	----	----
Texas:											
Dallas	11	1	1	0	4	8	0	1	0	0	61
Fort Worth	14	----	1	0	3	9	0	3	0	0	40
Galveston	3	----	0	0	0	0	0	0	0	0	12
Houston	12	----	0	0	5	1	1	4	1	0	64
San Antonio	7	----	0	0	4	0	0	3	0	0	55
Montana:											
Billings	0	----	0	0	1	14	1	0	0	0	7
Great Falls	0	----	1	0	1	1	0	0	0	6	8
Helena	0	----	0	0	0	1	0	0	1	0	3
Missoula	0	----	0	0	1	29	0	0	0	0	8
Idaho:											
Boise	1	----	0	0	2	4	0	0	0	0	7
Colorado:											
Colorado Springs	0	----	0	0	0	4	0	0	1	6	14
Denver	4	----	0	3	11	13	0	3	2	1	73
Pueblo	0	----	0	0	2	14	0	0	0	1	13
New Mexico:											
Albuquerque	0	----	0	0	2	3	0	4	0	5	18
Utah:											
Salt Lake City	0	----	0	3	5	40	0	0	0	1	34
Nevada:											
Reno	----	----	----	----	----	----	----	----	----	----	----
Washington:											
Seattle	0	----	0	1	8	13	1	7	0	1	117
Spokane	0	1	1	4	1	2	0	2	0	2	36
Tacoma	0	----	----	0	3	1	0	0	0	1	34
Oregon:											
Portland	0	----	1	19	5	12	0	3	0	1	80
Salem	0	----	----	0	----	1	0	----	0	1	----
California:											
Los Angeles	14	22	1	14	16	52	0	14	0	17	314
Sacramento	5	----	0	0	4	17	0	6	0	0	41
San Francisco	0	5	0	17	4	19	0	7	0	30	168

City reports for week ended Nov. 16, 1935—Continued

State and city	Meningococcus meningitis		Poliomyelitis cases
	Cases	Deaths	
New Hampshire:			
Nashua	0	0	1
Vermont:			
Barre	0	0	1
Massachusetts:			
Boston	0	0	8
Worcester	0	0	2
Rhode Island:			
Providence	0	1	1
New York:			
New York	3	1	11
Syracuse	0	1	5
New Jersey:			
Newark	0	0	3
Pennsylvania:			
Philadelphia	1	2	4
Ohio:			
Cincinnati	3	0	0
Indiana:			
Terre Haute	1	1	0
Illinois:			
Chicago	6	1	0
Springfield	1	1	1
Michigan:			
Detroit	4	0	3
Wisconsin:			
Kenosha	0	0	1
Minnesota:			
Minneapolis	1	0	1
Missouri:			
Kansas City	1	0	0
St. Louis	0	0	1
Kansas:			
Wichita	0	1	0
Maryland:			
Baltimore	2	0	0
District of Columbia:			
Washington	6	3	0
Georgia:			
Atlanta	1	0	0
Kentucky:			
Louisville	0	0	1
Tennessee:			
Knoxville	1	0	0
Memphis	1	2	0
Alabama:			
Birmingham	0	0	1
Louisiana:			
New Orleans	0	1	1
Colorado:			
Denver	0	0	2
New Mexico:			
Albuquerque	1	0	0
Oregon:			
Portland	2	0	2
California:			
Los Angeles	0	0	3
San Francisco	0	1	1

Epidemic encephalitis.—Cases: Toledo, 1; Chicago, 1; San Francisco, 1.
Pellagra.—Cases: Wilmington, N. C., 1; Winston-Salem, 1; Brunswick, 1; Miami, 1; Louisville, 2; New Orleans, 1; Dallas, 1.
Typhus fever.—Cases: Norfolk, 1; Savannah, 1; Montgomery, 1; Houston, 1; Los Angeles, 2.

FOREIGN AND INSULAR

CZECHOSLOVAKIA

Communicable diseases—September 1935.—During the month of September 1935, certain communicable diseases were reported in Czechoslovakia as follows:

Disease	Cases	Deaths	Disease	Cases	Deaths
Anthrax	8	1	Paratyphoid fever	22	----
Cerebrospinal meningitis	15	7	Poliomyelitis	48	1
Chicken pox	44	----	Puerperal fever	45	14
Diphtheria	2,084	106	Scarlet fever	2,310	21
Dysentery	132	16	Trachoma	96	----
Influenza	18	----	Typhoid fever	648	32
Lethargic encephalitis	6	2	Typhus fever	1	----
Malaria	311	----			

INDIA

Vital statistics—Quarter ended March 31, 1935.—Following are vital statistics for British India for the quarter ended March 31, 1935:

	Number	Rates per 1,000 population		Number	Rates per 1,000 population
Live births	2,220,973	32	Deaths from—Continued:		
Deaths	1,453,924	21	Fevers	819,587	12.0
Deaths from:			Plague	16,111	.2
Cholera	36,651	.5	Respiratory diseases	126,776	1.8
Dysentery and diarrhea	54,113	.8	Smallpox	23,668	.3

CHOLERA, PLAGUE, SMALLPOX, TYPHUS FEVER, AND YELLOW FEVER

NOTE.—A table giving current information of the world prevalence of quarantinable diseases appeared in the PUBLIC HEALTH REPORTS for November 29, 1935, pages 1701–1717. A similar cumulative table will appear in the PUBLIC HEALTH REPORTS to be issued December 27, 1935, and thereafter, at least for the time being, in the issue published on the last Friday of each month.

Plague

Ecuador.—A report dated November 13, 1935, states that the last case of plague reported in Duran, Ecuador, was on October 15, 1935, and the last plague-infected rat reported was on October 4, 1935. In Nobol, Ecuador, a small settlement located an hour and a half from Guayaquil, by automobile, 1 human case of plague was reported on October 20, 1935.

Egypt—Asyut.—During the week ended November 16, 1935, 1 death from plague was reported at Asyut, Egypt.

India—Punjab.—During the week ended November 9, 1935, 11 cases of plague with 5 deaths were reported at Punjab, India.

Iraq—Baghdad.—During the week ended November 9, 1935, 1 case of plague was reported at Baghdad, Iraq.

Yellow Fever

Colombia—Intendencia of Meta.—During the period September 29 to November 9, 1935, yellow fever was reported in the Intendencia of Meta, Colombia, as follows: Acacias, 2 cases; Restrepo, 2 cases, 2 deaths.

Ivory Coast—Abidjan.—On November 20, 1935, 1 case of yellow fever was reported at Abidjan, Ivory Coast.

Sudan (French)—Koutiala.—On November 20, 1935, 1 death from yellow fever was reported at Koutiala, French Sudan.

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